7/9/2012 – to be presented and discussed at ESC Meeting 11 July 2012

Vermont Endangered Species Committee

Recommendation to Secretary Markowitz on US Fish & Wildlife Lampricide Permit Applications for Missisquoi & Winooski Rivers for Fall 2012

I .Permit or Not to Permit? A statement as to whether the ESC recommends that the permit be granted or not granted.

The mortalities resulting from the lampricide applications are of great concern to the ESC, as repeatedly advised in the past. The citizen members of the ESC unanimously recommend that these permits not be approved. There are too many questions unanswered about consequences to other parts of the ecosystem and, more to our responsibility, to other species which are listed.

II. Permit Language/Conditions: Specific ESC recommendations for what should be stated in the permit itself (e.g., extend the distance to a relocation area, institute a mark-recapture study, shorten the permit period).

If the Secretary does approve these permit at the very least these restrictions should be applied as well as the conditions listed in part III:

- 1. Broadscale non-target surveys should include all Endangered & Threatened species potentially affected plus mudpuppy; all fatalities should be documented; and all specimens preserved for study.
- 2. Application should be made as late in the season as possible, to minimize mortality to the sturgeon.
- 3. Detailed reports of results and mortalities should be presented to the ESC.
- 4. Prior to consideration of this permit request, potential direct or indirect impacts to adult or young Spiny Softshell turtles should be addressed in the permit request along with post-treatment surveys for mortality of turtles. Collection of dead specimens for analysis should be included.
- 5. Mudpuppies:
 - non-target mortality surveys should cover as large a portion of the treated rivers as is possible and that any and all dead Mudpuppies be collected and stored in such a way as to allow future sexing and genetic analysis.
 - The needed research must be undertaken to show the current distribution and abundance of Mudpuppies in Vermont and performing the research to show the pre and post treatment population sizes of Mudpuppies in treated and untreated rivers in Vermont over a multiyear period.

After the many years of hearings and committee discussions regarding the use of lampricides, it is apparent that the current method of permit review wherein there are no discussions and reviewers review in isolation is difficult, inefficient, and of limited scope. The current method of review works well for simpler permit applications, but for permit applications dealing with large scale, complex circumstances ecosystem wide, the lack of direct conversations among reviewers

and the applicant has significant limitations. We request that in the future the ESC meet directly with the applicant for discussion.

Additional conditions recommended:

- 1. The applicant should be required to conduct annual monitoring surveys for all threatened and endangered species in all tributaries currently in the lampricide treatment rotation, and for tributaries being considered for future lampricide applications. The applicant should provide annual reports summarizing these monitoring efforts, including an analysis of the condition of each of these discrete threatened or endangered populations. This information will allow better-informed permit recommendations for future permit applications, and will also provide clear documentation of the condition of these protected populations over time. In the absence of plans to establish an adequate monitoring effort on rivers where these species occur, the ESC cannot support further chemical treatments for sea lamprey control.
- 2. If allowed to treat both rivers, the applicant should treat no sooner than September 24 and as late as possible to avoid significantly impacting sensitive young lake sturgeon. The treatment should be as late in the season as possible.
- 3. The primary application point (AP) on the Missisquoi River should be moved downstream to an area below prime stonecat habitat to the approximate area of the Swanton Town boat ramp approximately 250m downstream from the dam.
- 4. If the treatment is allowed on the Missisquoi River and the AP is located above the Swanton Dam, the applicant must not apply a lampricide at a concentration of more than 1.1XMLC.
- 5. If the treatment is allowed on the Missisquoi River and the AP is located above the Swanton Dam, all stonecats observed in the riffle section below the dam during the treatment that are significantly affected should be collected and placed into fresh river water and placed back into the river only after treatment is completed and the lampricide block has passed.
- 6. All American brook lampreys killed in either treatment should be retained for possible genetic work and submitted to Ken Cox of ANR and/or Dr. Donna Parrish at UVM.
- 7. The applicant should also conduct two post treatment surveys for American brook lampreys over areas along the shoreline where the bottom is visible over a section of river from immediately downstream from the primary AP to 5km downstream (corresponding with areas most likely to support American brook lamprey). Two post treatment surveys should also be completed for dead stonecats and should cover the entire visible area of hard bottom substrate between the AP and the Swanton boat launch on the west bank. The first survey should be conducted within 24 hours of treatment and the second, 48 hours following treatment.
- 8. A toxicity test be conducted on juvenile stonecats, and the results shared with the ESC.

III. Application Review: Points of concern and observations about the permit application itself and the proposed project.

COMMENTS FROM Bill Barnard, ESC VICE CHAIR, & PERMITS COMMITTEE MEMBER

I am responding to the E&T Species Taking Permit for the Winooski and Missisquoi Rivers. For the most part, my comments are similar for both river permit application. I suspect that I will have a minority opinion, so please share with the ESC if you feel inclined. I am going to make the suggestion that the Endangered Species Committee recommend that both permits be denied.

My reasons are two-fold. The applicant should be prepared to show that the application does not have an adverse effect on the populations of listed T&E species. We have repeatedly asked for pre-and post application surveys of these species and they were not presented to the ESC and its SAG groups as part of the current application.

I am confident that the application of TFM and !% niclosamide will have negligible effects on listed mussel species. Previous surveys and cage studies for the most part support the applicants assessment that the species is "not expected" to be impacted. The same is true for the Sand Darter. I am not convinced that the mortality for the Channel Darter can be described as "negligible". Mortality of this species has occurred in a previous treatment and the method of post treatment surveys for non-target species is less than complete.

In my opinion, the anticipated mortality of the American Brook Lamprey is unacceptable by the ESC. At the treatment level, it is almost certain that mortality of this listed species will exceed the 90% acknowledged by the applicant. How can that be considered a "negligible population effect"? There is no evidence that refugia populations above the application point in the Missisquoi and Sunderland Brook tributary in the Winooski will allow for recolonization of the impacted brook lamprey. This is supposition by the applicant based upon no research.

Likewise, I argue that a 10% mortality of Stonecat in the Missisquoi is unacceptable and reason for denial of that permit.

I recognize that my recommendation for denial of permit might not be accepted by the ESC or Secretary. Therefore, the recommendations of the Fish SAG should strongly recognized. Specifically, the need for the applicant to assure this committee in the future that the listed species are not being impacted. This evidence should be presented with subsequent applications for treatment. Failure to do so should result in denial of permit. The majority of Stonecats in the Missisquoi are in a riffle area below the application point. The Fish SAG and I feel that the application point be moved downstream of this rocky section. If the applicant cannot or will not, then, as a condition of the permit, there should be people in the water at the time of application with nets to catch stressed Stonecats and remove them to clean water.

The ESC has received these permits for a number of years. I believe the time has come to put a halt to lampricide treatments until it can be shown that there is no effect on the listed species we are charged to protect. I remind you that that is the applicant's responsibility. We have repeatedly expressed concern that his information is not available.

Statement after reviewing SAG-Fishes Recommendation

ANR needs to fish or cut bait (apologies for the aphorism, I couldn't resist) about doing the research on the populations of listed species at all the streams and rivers to receive lampricide this and future years. The SAG-Fish gives a clearly a damning report, and I agree with each and every point. Point by point by point the Fish SAG demonstrates the current willingness of the applicant and previous former applicant (Vermont Department of Fish and Wildlife) to make takings decisions based on no acceptable data regarding population status, extent of fatalities, recovery of individual organisms and populations, recruitment from other waters to replace takings losses, and more This has got to be resolved. It is worse than bad science, in many cases it is no science.

COMMENTS FROM IAN WORLEY, ESC & PERMITS COMMITTEE MEMBER

Winooski River application.

- 1. (a) Regarding mussels I concur with the comments by Peter Wimmer and echo his questions and concerns about the use of Niclosamide for the first time in the river.
- 2. (b) regarding mussels I appreciate the attention to the precision (and absolutely necessary) control of lampricide levels throughout the waters of its proposed use. Also, it is encouraging to note that there have been few mortalities found in post-application surveys. However, the strength of those data is very weak, considering the lack of pre-application numerical surveys and any direct assessment of long-term impacts on individual mussels. I would recommend continued research and innovation in these shortcomings to be pursued and/or support by the applicant and Vermont Fish and Wildlife.
- (c) Regarding mussels I am pleased to see some research underway on the effects of lampricide on the early life stages of mussels. Such work seems very important and has repeatedly been requested by the Endangered Species Committee.
- (d) Regarding fish I concur with the comments by Peter Wimmer and Bill Barnard about the timing of the application of the Lampricide and the effect on young Sturgeon.
- (e) Regarding fish, I would like to address section (3) of the takings permit application. It concerns the use of the term "negligible" in regard to the effect of lampricide on the three listed species. A typical definition of negligible is: "So small or unimportant as to be not worth considering; insignificant." The use of the term here in the permit application could be applied in two ways: it could mean "not of statistical significance," or it could mean "not of value." As currently used in section (3) I believe members of the Committee might disagree whether there will be a "negligible" effect or not. It is important to quantify these terms, in order to put them in a context suitable for the Committee to assess the potential risk and the abundance of the takings. In the case of Sturgeon Attachment 1 presents a lengthy description of a strategy to minimize Sturgeon mortality. I agree with the questions and issues by Peter Wimmer and would like to see the applicant's responses.

In the case of the Eastern Sand and Channel Darters I understand the discussion and conclusions by the applicant. In the case of American Brook Lamprey the determination of "negligible" hinges on

the Sunderland Brook population as a recruitment source for the high mortality expected in the Winooski is more directly discussed in terms of the population as a whole. There seems to me to be inadequate data and thus more risk in this than stated by the applicant.

Missisquoi River application.

I have read the assessments provided by Peter Wimmer and Jim Andrews. I have not received yet a copy of Rich Langdon's review. I may have additional comments once I receive Rich's thoughts.

- (a) Regarding mussels I concur with the comments by Peter Wimmer.
- (b) Regarding mussels I appreciate the attention to the precision (and absolutely necessary) control of lampricide levels throughout the waters of its proposed use. Also, it is encouraging to note that there have been few mortalities found in post-application surveys. However, the strength of those data is very weak, considering the lack of pre-application numerical surveys and any direct assessment of long-term impacts on individual mussels. I would recommend continued research and innovation in these shortcomings to be pursued and/or support by the applicant and Vermont Fish and Wildlife.
- (c) Regarding mussels I am pleased to see some research underway on the effects of lampricide on the early life stages of mussels. Such work seems very important and has repeatedly been requested by the Endangered Species Committee.
- (d) Regarding fish I concur with the comments by Peter Wimmer and Bill Barnard about the timing of the application of the Lampricide and the effect on young Sturgeon.
- (e) Regarding fish, I would like to address section (3) of the takings permit application. It concerns the use of the term "negligible" in regard to the effect of lampricide on the three listed species. A typical definition of negligible is: "So small or unimportant as to be not worth considering; insignificant." The use of the term here in the permit application could be applied in two ways: it could mean "not of statistical significance," or it could mean "not of value." As currently used in section (3) it would be hard for the ESC to agree that there will be a "negligible" effect.

In the case of Sturgeon (Attachment 1 page 22) "negligible" is expressed as "not suffer significant mortality". In section 12 of the application the term used is "minimal." It would be helpful to quantify expressly what would constitute "negligible," "not significant," and "minimal". Ideally these would be stated as some percentage of total population. This would give the Endangered Species Committee numbers of a kind similar to those used by the committee when evaluating the status of a species.

Without the numbers it is much more difficult for the committee to evaluate whether or not it agrees that the possible takings is "negligible" or "not significant."

In the case of Stonecat, 22 Stonecats were found dead following the 2008 lampricide application. Attachment 1 page 25 describes this result as causing "particular concern." I infer that this result was thus not "negligible." In the footnote for the species in section (3) of the application "negligible" appears defined as less than 10% mortality. I would like to see added to the discussion of Stonecats (Attachment 1 page 25) more quantitative documentation connecting the existing population numbers above and below the dam, the reference to 22 fatalities as being of "particular concern", the definition of 10% mortality as being "negligible", and the effect of a 10% mortality on the survivability of the population. Without this expanded detail, the Committee will not be able to make an assessment as to whether the lampricide application to the river will result in acceptable or unacceptable numbers of deaths.

In the case of American Brook Lamprey the determination of "negligible" is more directly discussed in terms of the population as a whole. I need to see Rich Langdon's report before offering further comments here.

FROM THE SAG-Invertebrates

Some Observations and Comments on the Permit Documents for the TFM and TFM/1%niclosamide Treatment of the Lamoille and Winooski Rivers for Sea Lamprey Control, Fall 2012

Submitted 6/21/12 H. Peter Wimmer, Chair of the Invertebrate SAG

General comments and observations pertaining to both rivers

A/ It appears that young Lake Sturgeon are particularly sensitive to TFM and the TFM/1% niclosamide mix. Great care must be exercised to protect the young fish from the size of yolk sac loss to approximately 70 mm TL to avoid high % kills.

Yolk sac sturgeon have greater resistance to TFM or TFM/ 1% niclosamide. Why might this be so? With the yolk sac present the young fish are getting nutrition directly from the yolk while once they loose the sac their nutrition comes from normal alimentation involving the circulatory system of the alimentary canal. Could TFM cause failure of the system or maybe the TFM is absorbed through the gut at a much greater rate than through the skin putting more of it in circulation where it has its physiological effects, primarily on the cytochrome oxidative pathway.

B/ According to test results, NOEC occurs at up to 1.5 x MLC of either treatment type so there would appear to be little danger of mussel kills at 1.0 x MLC. All the species that are either E or T except the fluted shell are at least marginally safe at this level.

C/ With this information in hand the treatment level for the Missisquoi will be 1.2 x MLC of TFM while the Winooski will be treated at 1.0 x MLC of either TFM or TFM/1% niclosamide for juvenile sturgeon protection. The 1.0 x MLC has been considered too low for 100% lamprey kill and consequently many Great Lake states and NY use 1.5 x MLC as their treatment level. This level is too high to confer safety on several T & E mussels and young sturgeon. The argument is made that older sturgeon survive treatment and their reproduction more than replaces the young killed by treatment, assuming that treatment will not be a yearly event.

Winooski River and use of Niclosamide or Baylucid

<u>A/ B</u>aylucid (niclosamide) is used in many areas of the world to control or eliminate water snails which feed on wetland crops and /or are intermediate hosts of a number of human flatworm parasites. It is considered to be a powerful molluscaside and could have unforeseen long term effects on mussels. It could also kill snails and some non-target mussels, glochidia on fish and newly released juvenile mussels.

The description of the 20% niclosamide 2 hour pre-treatment of the river is not clear. What will the actual concentration of niclosamide in terms of MLC be? We assume it won't be 20%! Is it being monitored during this preparatory "flush" of the river? Will the high performance liquid chromatography equipment be on site to perform the analysis of niclosamide to 1 ppb or will samples be sent to a lab for analysis and simply represent the levels present in the river "after the fact?" (Attachment 2, pg 1, item G). Does Niclosamide respond to pH, alkalinity, temperature & etc in a similar way as does TFM? If not, how will this be taken into account for maintaining a 1.0 x MLC for sea lamprey?

B/ What is the TFM level in terms of MLC likely to be since niclosamide approximately doubles the effectiveness of TFM?

C/ Isn't a 1.0 x 99% MLC considered to be borderline effective to attain a high % lamprey kill? Most Great Lake states and New York use an MLC of 1.5 because of its effectiveness in getting a high % sea lamprey kill. Vermont is using 1.0 x MLC of TFM/1% niclosamide for the treatment of the Winooski. This is being done in large part to protect juvenile sturgeon and some T & E mussels which may have low MLC levels but well above those for juvenile sturgeon. The states using 1.5 x MLC for lamprey control realize that juvenile sturgeon mortality will occur but indicate that if TFM treatment is not a yearly event, the reproductive age sturgeon will rapidly replace the TFM killed juveniles. The reduction in lamprey parasitism increases the health of the growing young and the reproductive individuals ultimately resulting in a greater sturgeon population in better health with higher reproductive rates. Should Vermont follow this practice?

C/ Why isn't the 4th column of the table, Item 3 of the "Application for E & T Species Taking Permit" filed in? The column is headed % of population to be collected/impacted. The expected mortality of the American Brook Lamprey is about 90% which is not a "negligible number yet the population effect in column is listed as negligible! Is this "population effect" to be discounted because of the population in Sunderland Brook which may be low enough that it will take a number of years to repopulate the Winooski? It would seem more honest to state that there will be at least a significant impact on the Winooski ABL population which will last several years?

Missisquoi River TFM Treatment

A/ The Northern Pike, particularly juveniles, is not listed as a species of concern this year but in 2008 there was considerable discussion of the welfare of this fish. What has changed, if any thing?

Additional StatmentWinooski/Missisquoi TFM/TFM-Niclosamide Treatment- Final Statement

H. Peter Wimmer, Chairman SAG Inverts, 7/1/12

It would appear that the Herps and Fish SAGS, and the ESC Permits Committee members Dr. Ian Worley and Dr. Bill Barnard recommend that both permits should be denied for numerous reasons. Dr. Barnard spells out the reasons in his email of 6/28/12. The SAG-Invertebrates support these evaluations of the permit applications. We are especially concerned about the use of Niclosamide in the Winooski and of the potential damage to the American Brook Lamprey populations in both streams and Stone Cat in the Missisquoi. Inadequately explored "refugia" as sources of repopulation can't be depended on.

However, since it is highly unlikely that the treatments will not occur, SAG Inverts recommends that pre- and post-treatment studies of the health and populations of all T & E species be carefully performed according to best scientific methods, noting signs of distress and population status (declining, growing or stable). These studies should occur at least yearly, if not more often, up to the date of the next proposed treatment of the rivers four years hence (applies to treatment of other streams as well). These study results should be made available to the ESC when the next lampricide permit for these and other streams is applied for.

A possible way of determining reproductive success of mussels might be to count the number of glochidia attached to known host fish (TFM killed or live caught) found in the stream on a yearly basis at the time when each mussel species is known to release glochidia. Up to the present the primary criterion for evaluating the effect of lampricide on mussels has been "live. dead or narcotized". This set of standards has typically been applied for up to a week after treatment. There have been a few studies following the survival of mussels after lampricide treatment for up to a year using the above mentioned criteria.

The impact of lampricide on the periphyton (aufwuchs) community has been poorly studied with Maki's 1976 artificial stream study being the one still quoted most often. New studies are needed of the aufwuchs and in-fauna (those organisms living below the surface of the stream bottom) in streams and in the lab. It would be useful to know the effect of lampricide in these communities since they contribute significantly to the particulate organic particles in the water column upon which both lamprey larvae and mussels feed on.

Niclosamide degrades fairly rapidly in clear water but in turbid water as found in the Winooski it binds to clay silt particles which settle to the bottom where mussels may ingest them long after it has cleared from the water column. This extended exposure to ingested sub-lethal niclosamide may result in decreased health and early death. Niclosamide has been shown to interfere with the

function of the cytochrome oxidase system which depresses metabolism in snails, helminth parasite adults and their cercaria. Long term sub-lethal exposure has been shown to result in decreased longevity and respiration of snails. Is it unreasonable to expect that it may have the same effect on mussels?

Niclosamide is approximately 43 times as toxic to fish as TFM hence demanding special vigilance during its application.

Recommendations of the Scientific Advisory Group for Fishes on the Threatened and Endangered Species Takings Permit submitted by the US Fish and Wildlife Service for the application of Chemicals to control Sea lamprey in the Winooski and Missisquoi rivers in 2012.

Submitted by Rich Langdon, Chair Fish SAG, June 25, 2012

The Winooski River is proposed to be treated once at **1.0XMLC** with TFM or a mix of TFM and niclosamide for a period of 12-14 hours. MLC is the minimum lethal concentration required to kill 99.9% of sea lampreys in 9 hours. This term is used because the amount of lampricide needed to successfully treat varies between locations and time. The proposed treatment date is between September 3 and December 1 of 2012. The Missisquoi River is proposed to be treated once with TFM at a concentration of **1.2XMLC** for 12-17 hours between the same dates as the Winooski River.

Since the T&E permit applications for these two rivers entail many of the same fish species, our comments and recommendations for the two applications will be combined and presented by fish species. The first section briefly presents toxicity information for the listed species and the second will summarize our comments and recommendations.

Listed Species Toxicity

Eastern Sand Darter-state threatened. Occurrences: - Winooski, Poultney, Lamoille, and Missisquoi rivers. Toxicity tests on the eastern sand darter indicate a maximum no-effect lampricide concentration (NOEC) of **1.3-1.4 X MLC**. Only one post treatment mortality (Winooski at **1.0MLC**) of this species has been reported in the last eight lampricide treatments.

Channel Darter-state endangered. Occurrences: Poultney, LaPlatte, and Winooski rivers. Toxicity tests on the channel darter showed an NOEC of **1.2 X MLC**. Post treatment surveys on four treatments on the Poultney River (0.8-1.3MLC) reported no mortalities while four were reported over the two treatments in the Winooski River (both at 1.0MLC).

Lake Sturgeon-State endangered. Occurrences: Otter Creek and Winooski, Lamoille and Missisquoi rivers. Toxicity tests show very young sturgeon to be highly sensitive to TFM and the TFM-Niclosamide mixture. Resistance substantially increases however, during the first year of growth going from **0.45XMLC** soon after hatch to **1.46XMLC** at the end of the first growing season. Only one post treatment mortality has been reported (Winooski <u>River</u>, 2004, at **1.0XMLC** TFM). Caged juvenile lake sturgeon showed no mortality during the 2004 Winooski River treatment.

Stonecat- state endangered. Occurrences: La Platte and Missisquoi rivers and Hungerford Brook (tributary to Missisquoi). Stonecats are sensitive to TFM and most probably the TFM- niclosamide mixture. Caged stonecats in a Great Lakes tributary where TFM concentrations ranged between **1.1** and **1.2 XMLC** experienced 9% mortality. More recently, a toxicity test was conducted by the applicant in 2011. The test was replicated three times on different dates at water temperatures of 28, 22 and 8 degrees C. The 12-hour no effect concentrations were **0.9**, **1.2**, and **1.3XMLC** respectively. The lowest effect concentration ranged from **1.2 to 1.5XMLC** with mortality ranging from 40-60%. The 2008 Missisquoi River TFM treatment resulted in 22 stonecat mortalities. However during the early stage of treatment, 36 affected stonecats were removed from the treatment area and would have likely died if not removed and placed upstream above the application point. This lampricide-induced stress was likely increased due to a malfunction in the delivery line early in the treatment which caused an increased TFM concentration along the bank where these fish were collected.

American Brook Lamprey-state threatened. Occurrences: Winooski and Missisquoi rivers, Sunderland, Trout, Stonebridge, Kelly, and Youngman brooks. The American brook lamprey is slightly less sensitive to TFM and the TFM- niclosamide mix than are sea lamprey. During the 2004 Winooski TFM treatment at 1.0MLC, caged American brook lamprey suffered an 82% mortality rate. From New York Champlain tributaries, cage and population estimates show similar results where sea lamprey mortality indicated a successful treatment. Toxicity tests show that a 9- hour exposure of TFM that would kill nearly 100% of sea lamprey (1.0XMLC) would also cause 50-60% mortality in the brook lamprey. An exposure of 12 hour at 1.0XMLC however would cause 100% mortality brook lamprey. Prior to the 2004 Winooski treatment, American brook lamprey were collected during sea lamprey pre- treatment monitoring. None were collected during the 2008 pretreatment survey.

Comments

Because these five species are state listed and has primary distributions that overlap that of the sea lamprey, our discussion will reflect a program-level perspective rather than limiting our scope to the two rivers under consideration. Toxicity information for the two darter species implies a risk of mortality from lampricide treatments that have ranged from 1.0-1.3XMLC. The treatment history for these two darter species however has shown only minimal mortality. FSAG however has concerns that the methods of assessing nontarget mortality for all species provide only a coarse estimate of losses. Post treatment observations only cover the visible portion of the river bottom, and population estimates have very wide confidence bands implying a very low level of precision. While the prior applicant conducted a pre and post treatment channel darter population survey of the 2007 Poultney River treatment, the results were equivocal and reflected difficulties in sampling these two small fishes. A demonstration of the Missouri benthic trawl net last year showed remarkable efficiency in capturing darters in Vermont rivers that are treated with lampricide. Since FSAG believes that monitoring these species in treated rivers is a necessary element for insuring proper protection, we believe the use of the benthic trawl may remove a significant obstacle to effective population monitoring of these two species in the future - if further study verifies its potential effects on other organisms are minimal. In the absence of plans to establish an adequate monitoring effort on rivers where these species occur, FSAG cannot support further chemical treatments for sea lamprey control.

Older juvenile lake sturgeon should be sufficiently resistant to lampricide to avoid effects of a treatment at the proposed concentration. FSAG believes then, that if a treatment takes place on either or both rivers, that the treatments take place on or after September 24 rather than September 3 as proposed.

Because of reported stonecat mortalities resulting from the last Missisquoi treatment as well as the recent toxicity test and the fact that this species only occurs in two rivers in Vermont, our level of concern is high for this species. The proposed treatment level (1.2XMLC) is at the toxicity tests-calculated mean maximum allowable treatment concentration (MATC), which is the mean of the NOEC and LOEC. The MATC calculated in this manner may be inaccurate because it assumes that the dose response relationship between the NOEC and the LOEC is linear. If it is not linear, mortalities could be as low as 0 and as high as 60%. There is currently no regular population monitoring of stonecat populations. However a new population study has recently been undertaken by UVM and ANR researchers. FSAG believes a long term monitoring effort on the Missisquoi River is necessary in order for this group to support further permit applications for sea lamprey control. We also question the placement of the application point ABOVE the Swanton Dam. Placing the AP at or downstream from the public boat ramp in Swanton would avoid most if not all of the stonecat habitat in that section of the river and protect this species from impact.

The American brook and silver lampreys are the most sensitive nontarget species in this program. While numbers of silver lamprey appear to consistently rebound following treatments, small American brook population sizes in the Winooski and Missisquoi rivers prevent accurate monitoring of population conditions. Consequently, population resilience of this brook lamprey is unknown. Current efforts to monitor numbers of American brook lamprey have proven to be insufficiently intensive to produce meaningful data. FSAG members believe that monitoring should include increased effort and be conducted on an annual basis so that reasonable conclusions could be drawn on the impacts to this species. FSAG does not agree with the applicant's assertion that Sunderland Brook, and Kelly and Hungerford brooks provide recolonization sources for the Winooski and the Missisquoi river populations respectively. We know of no available literature or other information that supports this hypothesis. The same dearth of information on movement exists for the stonecat as well. Without rigorous population monitoring of brook lamprey numbers FSAG cannot support further chemical treatments. In addition, current toxicity information and cage tests on this species indicate a clearly excessive mortality rate that is unacceptable to this SAG.

Recommendations

- 1. We acknowledge the high likelihood of chemical treatments to proceed on these two rivers this year. As such, we strongly urge that the applicant be required to conduct annual monitoring surveys for threatened and endangered species in all tributaries currently in the lampricide treatment rotation, and for tributaries being considered for future lampricide applications. The applicant should provide annual reports summarizing these monitoring efforts, including an analysis of the condition of each of these discrete threatened or endangered populations. This information will allow the FSAG to provide better-informed permit recommendations for future permit applications, and will also provide clear documentation of the condition of these protected populations over time.
- 2. If allowed to treat both rivers, the applicant should treat no sooner than September 24 or later to avoid significantly impacting sensitive young lake sturgeon.
- 3. The primary application point (AP) on the Missisquoi River should be moved downstream to an area below prime stonecat habitat to the approximate area of the Swanton Town boat ramp approximately 250m downstream from the dam.
- 4. If the treatment is allowed on the Missisquoi River and the AP is located above the Swanton Dam, the applicant must not apply a lampricide at a concentration of more than 1.1XMLC.

- 5. If the treatment is allowed on the Missisquoi River and the AP is located above the Swanton Dam, all stonecats observed in the riffle section below the dam during the treatment that are significantly affected should be collected and placed into fresh river water and placed back into the river only after treatment is completed and the lampricide block has passed.
- 6. All American brook lampreys killed in either treatment should be retained for possible genetic work and submitted to Ken Cox of ANR or Dr. Donna Parrish at UVM.
- 7. The applicant should also conduct two post treatment surveys for American brook lampreys over areas along the shoreline where the bottom is visible over a section of river from immediately downstream from the primary AP to 5km downstream (corresponding with areas most likely to support American brook lamprey). Two post treatment surveys should also be completed for dead stonecats and should cover the entire visible area of hard bottom substrate between the AP and the Swanton boat launch on the west bank. The first survey should be conducted within 24 hours of treatment and the second, 48 hours following treatment.
- 8. The FSAG recommends that a toxicity test be conducted on juvenile stonecats.

Input from the Reptile and Amphibian Scientific Advisory Group on the 2012 TFM Permit Applications – Jim Andrews, Chair June 2012

This input is based on discussions in many previous Herp SAG meetings over the last decade. The last set of "other suggested permit conditions" is entirely new and written by me as chair without input from the SAG. The SAG-Herps does not support the granting of these permits.

Mudpuppies

Since Mudpuppies are not currently listed, the argument has been made by the applicant that concerns about this species should be addressed through the Aquatic Nuisance Control Permit process. As you know, the Herp SAG has no authority to make recommendations through that process, nor are there groups of taxonomic experts set up to give input through that process, consequently this is our only opportunity for input as a group on this species and we provide the following recommendation.

Although Mudpuppies are not currently listed, we have twice proposed this species for listing as threatened and it is our consensus that were it not for Vermont Fish and Wildlife's departmental position of support for the TFM program and resulting lack of support for listing Mudpuppy, Mudpuppies would be and should be currently listed. Mudpuppy mortality during the last treatment

of the Lamoille River was at a minimum 528 individuals (twenty times previous levels) and based on the percentage of the treated portion of the river surveyed (5%) actual direct mortality could have been in the thousands of individuals. Recent genetic work completed in Dr. William Kilpatrick's lab at UVM has shown that the Mudpuppy in the Connecticut River is not closely related to any other native Mudpuppy populations in the northeastern United States. Consequently, the distribution of our native lamprey is limited to the same range that is treated by lampricides and Mudpuppies have suffered significant mortality during those treatments. Mudpuppies appear to have been entirely extirpated from Lewis Creek or at the very least reduced to a population size so small that it can no longer be detected.

Follow up studies at UVM show that Mudpuppies are still in the Lamoille River. This is of course good news. However, this should not be taken as justification for continued use of lampricides. Our standard for rare species with limited distribution should not be, "did some of the population survive the treatment". Instead, in our recovery plans we do everything we can to enhance rare species, remove sources of mortality, and grow populations. Aside from trying to enhance the population, we still have the question of whether or not a population of a long-lived species (20-30 years) can withstand regular mortality of all age-classes (particularly adults) on a sustained basis. In evaluating past mortality of Mudpuppies, Dr. Kilpatrick and I agree with former NY DEC Herpetologist Al Breisch that in addition to mortality of adults, TFM treatments seem to have almost entirely removed certain young age-class Mudpuppies from some NY populations in previous treatments. This increased toxicity to younger age-classes is also supported by other studies. Based on the mortality from the relatively recent Lamoille River treatment, TFM impacts on Mudpuppies have not been minimized or reduced over the years.

I assume that the new applicant (USF&W) now has taken on the responsibilities that were required of the past applicant. One of those responsibilities that has yet to be met is to gather and provide information regarding the distribution and abundance of Mudpuppies in other streams (and Lake Champlain) in Vermont. As always, we feel the burden of proof that periodic TFM treatments is sustainable with regard to Vermont Mudpuppy populations, should be on the applicant. We have not yet seen the studies that show healthy pre- and post-treatment populations of Mudpuppies, let along long-term studies.

As always, we continue to be concerned about potential sub-lethal, long-term, and synergistic effects of the regular use of these chemicals. No studies have ever been provided to us that investigate these types of impacts of TFM despite numerous examples of these sorts of impacts from other commonly used biocides.

Consequently, we do not recommend the approval of these two permit requests. If permission is granted, we recommend the following conditions:

- 1. that non-target mortality surveys cover as large a portion of the treated rivers as is possible and that any and all dead Mudpuppies be collected and stored in such a way as to allow future sexing and genetic analysis.
- 2. performing the needed research to show the current distribution and abundance of Mudpuppies in Vermont and performing the research to show the pre and post treatment

population sizes of Mudpuppies in treated and untreated rivers in Vermont over a multiyear period.

Spiny Softshell Turtles

The following is adapted from the input regarding Spiny Softshells that the Herp SAG presented in response to the 2009 request for treatment of the Lamoille River and it is still **pertinent for the Missisquoi application**. Although there has been no direct mortality of turtles reported from prior TFM treatments, the presence of the species should be acknowledged and the potential risks addressed in the Missisquoi River permit request. Synergistic, sub-lethal, and indirect impacts have never been addressed for this species.

The Missisquoi River serves as important foraging, basking, and overwintering habitat for the state-threatened Spiny Softshell turtle (*Apalone spinifera*). We have concerns about the potential impact either on the turtle directly or indirectly over time through synergistic, or other long-term sub-lethal impacts. We have not seen the type of controlled long-term study that would show it has no impact. Impacts on young turtles and indirect sub-lethal or long-term impacts are a concern.

Spiny Softshells have shown sensitivity to some aquatic biocides (rotenone) and as a result of its thin leathery skin may be more sensitive to biocides in general than other turtles. Here is a quote from Pam Bartholomew at Michigan State;

"Because A. spinifera respire aquatically with pharyngeal gill slits and cloacae, they are vulnerable to Rotenone, a chemical that is used to kill unwanted fish. Rotenone hinders oxygen absorption and many softshell turtles are now gone from Rotenone contaminated waters in the Great Lakes."

A separate article on Red-Eared Sliders (not native) also supports concerns about pesticide impacts on turtles. Our Painted Turtles (*Chrysemys picta*) and Map Turtles (*Graptemys geographica*) are more closely related to the sliders than the Spiny Softshell and both are found in the Missisquoi River.

Embryonic exposure to low-dose pesticides: effects on growth rate in the hatchling red-eared slider turtle.

Willingham E. 2001. Division of Biological Sciences, University of Texas, Austin 78759, USA. ewillingham@austin.rr.com, Journal of Toxicology and Environmental Health A 2001 Oct 12;64

In the red-eared slider turtle, pesticides can alter expected sex outcomes, a major step in the inferred pathway of sex determination, and hatchling steroid physiology. Changes such as these can profoundly affect an organism's fitness. Other potential markers for effects on fitness include hatchling mass, hatchling use of maternal stores (residual yolk), and especially early hatchling growth rates. In the current study, red-eared slider turtles were exposed during embryogenesis to one of three compounds-chlordane, trans-Nonachlor, or p,p'-DDE-all of which affect sex determination in this species. Turtles were weighed at hatching, after a 28-d fasting period, and after 14 d of ad libitum feeding. All three

compounds had some population-wide effects on changes in mass from time point to time point when compared to controls. From hatching to the end of the 28-d fast, turtles exposed in the egg to the mid-range doses of trans-Nonachlor and of p,p'-DDE lost mass and underwent a change in mass significantly different from controls. Additionally, turtles exposed to the two higher doses of trans-Nonachlor and the mid-range dose of chlordane grew significantly more than controls after 14 d of ad libitum feeding. These results point to a role for pesticides in endocrine disruption that extends beyond sex determination and sex development.

As pertains to Spiny Softshells the HerpSAG recommends that the permit be denied. However, if the permits are granted, the RASAG recommends:

1. That prior to consideration of this permit request, potential direct or indirect impacts to adult or young Spiny Softshell turtles be addressed in the permit request along with post-treatment surveys for mortality of turtles. Collection of dead specimens for analysis should be included.